International Journal of Science and Research (IJSR)
ISSN: 2319-7064
Impact Factor 2025: 7.089

An Observational Study Evaluating the Role of Neutrophil-Lymphocyte Ratio and Platelet-Lymphocyte Ratio in Assessing Disease Activity in Ulcerative Colitis at a Tertiary Care Centre

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Abstract: ***Ulcerative colitis (UC) is a chronic inflammatory bowel disease characterized by relapsing and remitting mucosal inflammation. Endoscopy remains the gold standard for assessing disease activity but is invasive and less suitable for frequent monitoring. This observational study evaluated the role of neutrophil-lymphocyte ratio (NLR) and platelet-lymphocyte ratio (PLR) as non-invasive biomarkers of disease activity in UC. Forty-five clinicopathologically confirmed UC patients and 45 healthy controls were enrolled. Cases were classified into mild to moderate and severe disease according to the Truelove and Witts criteria. Hematological parameters, NLR, and PLR were calculated and compared between groups. Both NLR and PLR were significantly higher in UC patients than in controls and increased progressively with disease severity. ROC analysis demonstrated good diagnostic performance for both markers, with NLR showing superior sensitivity and diagnostic accuracy. The findings suggest that NLR and PLR are simple, inexpensive, and readily available biomarkers that may serve as useful adjuncts for assessing disease activity in ulcerative colitis, particularly where endoscopic evaluation is not readily available.***

Keywords: Ulcerative Colitis; Neutrophil-Lymphocyte Ratio; Platelet-Lymphocyte Ratio; Disease Activity; Inflammatory Bowel Disease; Biomarkers

1. Introduction

Ulcerative Colitis (UC), a type of Inflammatory Bowel Disease (IBD), is a chronic, idiopathic inflammatory disease affecting the colon, characterised by relapsing and remitting mucosal inflammation, starting in the rectum and extending to proximal segments of the colon [1]. UC has a bimodal age distribution with peak incidence in the second or third decades and a second peak between 50 and 80 years [2]. Males and females are equally affected. The pathogenesis is multifactorial, involving genetic predisposition, epithelial barrier defects, dysregulated immune responses, and environmental factors. Both innate and adaptive immunity are involved in pathogenesis [1]. Most patients present with bloody diarrhea, with or without mucus, abdominal pain, malaise, weight loss, and fever [3]. Endoscopy remains the gold standard for the diagnosis and assessment of disease activity in patients with UC. However, its routine use is limited by several factors, including its invasive nature, reduced feasibility for long-term monitoring, higher cost, and inter-observer variability in interpretation [4]. Moreover, endoscopy may not be available in all centers and has a propensity for complications in active colitis. Compared with endoscopy, non-invasive tests, such as erythrocyte sedimentation rate (ESR), white blood cell (WBC) count, platelet count, and levels of C-reactive protein (CRP), acid glycoprotein, and albumin are being recognized as important markers for initial diagnosis and disease activity detection. Since these parameters are not specific for UC disease activity, adjunctive use of additional serum markers is required to monitor disease activity [5]. Faecal calprotectin (FC) is a marker of gut inflammation and shows a good correlation with endoscopic inflammation in UC [6]. However, most patients show reluctance to stool investigations. Therefore, there is an unmet clinical need to identify biomarkers that can replace the need for an endoscopy to assess mucosal disease activity. A number of studies have been performed to study the role of neutrophil to lymphocyte and platelet to lymphocyte ratio in assessing severity of ulcerative colitis for effective follow up and management of ulcerative colitis. These studies have demonstrated a positive correlation between NLR and PLR ratios and disease activity in ulcerative colitis.

The neutrophil to lymphocyte ratio (NLR) and platelet to lymphocyte ratio (PLR) can be easily calculated from the complete blood count (CBC) and are simpler and less expensive biomarkers [7]. The study, therefore, aims to evaluate the usefulness of NLR and PLR as disease activity biomarkers in Ulcerative Colitis.

2. Materials and Methods

45 clinicopathologically confirmed cases of Ulcerative Colitis and an equal number of healthy controls were enrolled in the study. Patients with other known inflammatory conditions, atherosclerotic coronary disease, hepatosplenic disease, autoimmune disease, malignancies, pregnant and lactating mothers and patients with an identified active focus of infection were excluded from the study. Cases were clinically classified into mild to moderate and severe cases of Ulcerative Colitis based on Truelove and Witts Criteria for Severity of Ulcerative Colitis. EDTA anticoagulated venous blood samples were collected from both cases and controls and samples were processed using automated haematology analyser- Sysmex XN-1000.

The patient's age, gender, disease activity according to Truelove and Witts Criteria, haematological parameters, Neutrophil- Lymphocyte Ratio (NLR) and Platelet-Lymphocyte Ratio (PLR) were recorded. NLR and PLR were calculated by dividing the absolute neutrophil count by

absolute lymphocyte count and dividing the absolute platelet count by the absolute lymphocyte count respectively for both cases and controls. The NLR and PLR were assessed for mild disease activity and severe disease activity groups of Ulcerative Colitis as well as for healthy controls to evaluate utility of these as non-invasive biomarkers to predict disease severity.

3. Statistical Analysis

Data was entered into Microsoft Excel (Windows 10; Version 2021) and analyses were done using the Statistical Package for Social Sciences (SPSS) for Windows software (version 27.0; SPSS Inc, Chicago). Descriptive statistics such as mean and standard deviation (SD) for continuous variables, frequencies and percentages were calculated for categorical variables. The Chi-Square test was used to find association between categorical variables. Unpaired t test was used to compare mean of hematological parameters between cases and controls and between mild to moderate cases and severe cases. Sensitivity, specificity and predictive accuracy were calculated for NLR, PLR. The level of significance was set at 0.05. ROC (Receiver operating characteristics) curve was drawn for NLR and PLR and optimal cut- offs calculated between cases and controls and for mild to moderate cases and severe cases.

4. Results

45 cases of Ulcerative Colitis and 45 healthy controls were enrolled in the study. The following results were obtained.

The majority of cases were in the age group of 41–50 years (26.7%), followed by ≤20 years (17.8%), 21–30 years (17.8%), and 51–60 years (17.8%). The overall mean age of cases was 38.04 ± 15.87 years, while that of controls was 39.44 ± 14.07 years. Age did not act as a confounding variable in the study.

Among cases, 53.3% were males and 46.7% were females. In the control group, females constituted 55.6% while males accounted for 44.4%. Both groups were comparable in terms of gender distribution, suggesting that gender was unlikely to influence the study outcomes.

Ulcerative Colitis cases were divided into 2 groups- Mild to Moderate cases and Severe Cases- based on Truelove and Witts Criteria of Severity for Ulcerative Colitis.

Table 1: Distribution of Study Subjects according to the Disease Activity (N = 45)

Disease Activity	No.	Percent
Mild to moderate	26	57.8
Severe	19	42.2

Out of 45 cases, 57.8% had mild to moderate disease activity, while 42.2% had severe disease activity allowing meaningful comparison of both groups.

Table 2: Comparison of Haematological Parameters between Cases and Controls (N=90)

Parameter	Group		P Value
	Cases (n=45) Mean (SD)	Controls (n=45) Mean (SD)	
HB	10.67 (1.56)	13.19 (1.49)	<0.001*
TLC	10828 (2887)	7357 (2069)	<0.001*
Neutrophils	8039 (2919)	4456 (1394)	<0.001*
Lymphocytes	2089 (603)	2182 (843)	0.549
Platelet	4.459 (1.173)	2.642 (0.672)	<0.001*
NLR	4.305 (2.430)	2.222 (0.818)	<0.001*
PLR	237.34 (113.20)	134.51 (49.40)	<0.001*
Unpaired t Test, P Value * Significant			

Hemoglobin levels were found to be significantly lower, Total leukocyte count (TLC), absolute neutrophil count and platelet count were significantly higher in cases when compared to controls with high statistical significance (p<.001). Neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) were significantly increased among cases (p< .001), further supporting systemic inflammation in diseased individuals. However, lymphocyte counts did not differ significantly between groups (p = 0.549).

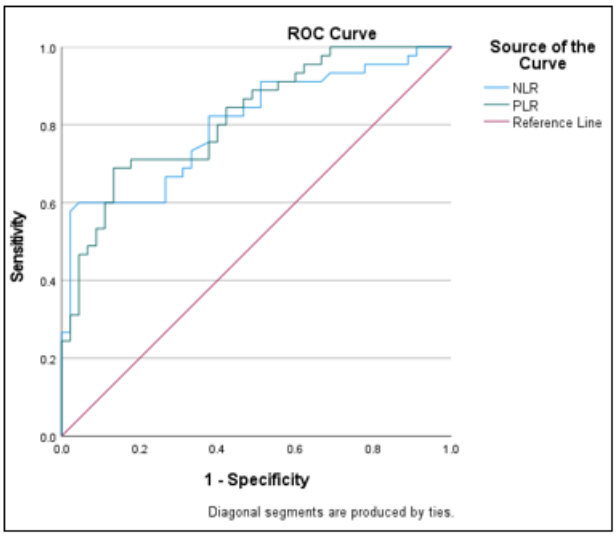
Table 3: Comparison of Haematological Parameters with Disease Severity (N=90)

Parameter	Group			P Value
	Mild to Moderate (n=26) Mean (SD)	Severe (n=19) Mean (SD)	Controls (n=45) Mean (SD)	
HB	11.59 (1.33)	9.40 (0.75)	13.19 (1.49)	<0.001*
TLC	8926 (2136)	13431 (1289)	7357 (2069)	<0.001*
Neutrophils	5995 (1782)	10837 (1454)	4456 (1394)	<0.001*
Lymphocytes	2301 (624)	1798 (440)	2182 (843)	0.060
Platelet	3.91 (1.10)	5.20 (0.81)	2.642 (0.672)	<0.001*
NLR	2.725 (0.898)	6.468 (2.183)	2.222 (0.818)	<0.001*
PLR	188.20 (96.89)	304.58 (100.12)	134.51 (49.40)	<0.001*
ANOVA, P Value * Significant				

Hemoglobin levels progressively decreased with increasing disease severity. TLC, neutrophil count, platelet count, NLR, and PLR showed a progressive and statistically significant increase from controls to mild/moderate cases to severe cases (p<.001). Although lymphocyte count was lower in severe cases compared to mild cases, the difference did not reach statistical significance (p = 0.060).

In a study conducted by Akpinar et al. the mean WBC, neutrophil, and platelet counts in the active endoscopic disease group were higher compared with those in the other groups (remission and controls), and these were higher in the patients in remission compared with the controls [6].

ROC Curve of NLR and PLR for Cases versus Controls:



Test Result Variable(s)	AUC	SE	P Value	95% Confidence Interval	
				Lower Bound	Upper Bound
NLR	0.803	0.046	<0.001*	0.712	0.894
PLR	0.819	0.043	<0.001*	0.734	0.904

Table 4.1: Sensitivity, Specificity and Predictive Value of NLR and PLR for Cases versus Controls (N=90)

Parameter	Cut-off	Sensitivity	Specificity	PPV	NPV
NLR	2.275	82.2	62.2	68.5	77.5
PLR	140.30	84.4	57.8	66.7	78.8

Table 4.2: Diagnostic performance of NLR and PLR for Cases versus Controls (N=90)

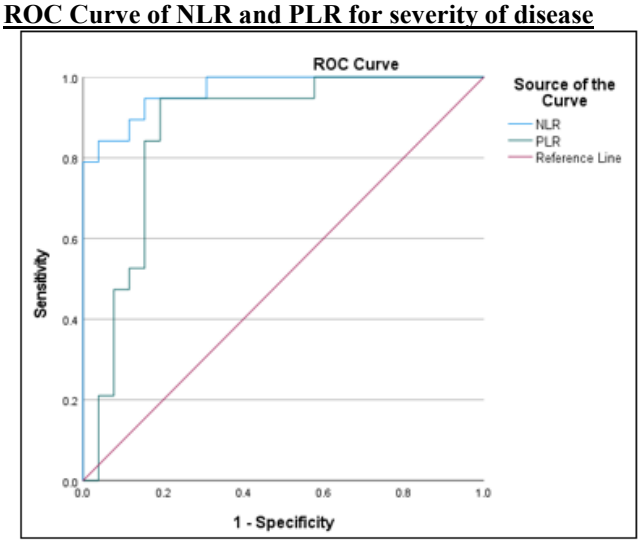
Parameter	Diagnostic Accuracy	Positive Likelihood Ratio	Negative Likelihood Ratio
NLR	72.2	2.17	0.29
PLR	71.1	2.0	0.27

A cut off of 2.275 was calculated for NLR from ROC analysis for differentiating cases versus controls. At this cut off, the NLR has a good sensitivity of 82.2%, but a moderate specificity of 62.2% with a diagnostic accuracy of 72.2%. PLR had a mean value of 237.34 (113.20) among cases. ROC analysis gave a cut off of 140.30 for PLR between cases and controls. At this cut off, PLR had sensitivity of 84.4% and a specificity of 57.8%. Overall, both parameters demonstrated comparable diagnostic performance, with PLR showing slightly higher sensitivity and NLR showing marginally higher specificity and diagnostic accuracy.

The study performed by Jeong et al. revealed a cutoff of 2.26 for NLR (sensitivity 54.2%; specificity 90.6%; LR+ 5.778) and 179.8 for PLR (sensitivity 35.4%; specificity 90.6%; LR+ 3.78) between cases and healthy controls. The AUC for NLR was higher compared to that of PLR indicating that NLR was more significant than PLR for diagnosing UC [7].

The cut offs are similar to our cut offs of 2.275 for NLR and 140.34 for PLR. However, our study shows higher sensitivities but lower specificities for both NLR and PLR for differentiating cases from healthy controls. In addition, the cutoff for PLR is slightly lower in our study. Overall, NLR was more significant than PLR for diagnosing UC.

ROC Curve of NLR and PLR for severity of disease



Test Result Variable(s)	AUC	SE	P Value	95% Confidence Interval	
				Lower Bound	Upper Bound
NLR	0.968	0.022	<0.001*	0.924	1.000
PLR	0.866	0.058	<0.001*	0.753	0.979

Table 5.1: Sensitivity, Specificity and Predictive Value of NLR and PLR for Severity of Cases (N=45)

Parameter	Cut-off	Sensitivity	Specificity	PPV	NPV
NLR	3.08	100.0	69.2	70.4	100.0
PLR	190.27	94.7	73.1	72.0	95.0

Table 5.2: Diagnostic Performance of NLR and PLR for Severity of Cases (N=45)

Parameter	Diagnostic Accuracy	Positive Likelihood Ratio	Negative Likelihood Ratio
NLR	82.2	3.25	0.00
PLR	82.2	3.52	0.07

In the present study, the cut off of NLR and PLR to differentiate between mild to moderate and severe disease activity were found to be 3.08 and 190.27 respectively. The sensitivities for NLR versus PLR at the specific cut off values are 100% and 94.7%; specificity 69.2% and 73.1%; PPV 70.4% and 72.0%; NPV 77.5% and 95%; Diagnostic accuracy 82.2% and 82.2%; LR+ 3.25 and 3.52; LR- 0.00 and 0.07 respectively. These results indicate that both NLR and PLR demonstrated good diagnostic performance in predicting disease severity, with NLR showing perfect sensitivity, whereas PLR demonstrated slightly higher specificity and positive likelihood ratio.

Jeong et al. demonstrated an optimal cutoff of 3.44 and 175.9 for NLR and PLR respectively to differentiate mild to moderate and severe case (sensitivity, 63.6% vs. 90.9%; specificity, 81.1% vs. 78.4%; positive likelihood ratio, 3.364 vs. 4.205; AUC, 0.714 vs. 0.897). PLR had the highest AUC and positive likelihood ratio in the study [7].

Our study revealed similar cutoffs for NLR and PLR. However, our study demonstrated that NLR and PLR had a higher sensitivity but a lower specificity when compared to the study of Jeong et al. Also, our study shows that NLR had a higher AUC and higher sensitivity when compared to PLR

in assessing severity of cases of UC which is contrasting to the results obtained by Jeong et al.

Feng et al. further emphasised the role of NLR and PLR as non-invasive markers in reflecting disease activity in UC. The receiver-operating characteristic (ROC) analysis revealed the optimal cutoff of NLR for active UC as 2.19, with sensitivity and specificity of 78.8 and 65%, respectively. For PLR, the best cut-off value was 147.96, with sensitivity and specificity of 58.3 and 75%, respectively, concluding that PLR and NLR were elevated in patients with active UC as compared with patients in remission [8].

In the study performed by Fidan K et al. the NLR of the active disease patients was significantly higher than that of those in remission. The ROC cut-off value for NLR to discriminate an active phase in UC patients was ≥2.2 (sensitivity: 62%; specificity: 70%) The cut-off value for PLR to discriminate active UC was calculated to be ≥133.87 using ROC analysis (sensitivity: 63%; specificity: 68%) [5].

A study by Demir et al. revealed that NLR values of the active UC group were elevated compared with those of the patients with inactive UC and the controls. However, ROC analysis revealed 2.39 as the optimum NLR cut-off value for active UC with a sensitivity of 48.6 and specificity of 77.5%. Thus, they concluded that NLR has low sensitivity and specificity rates in determining active UC, [9]. Our study findings are contradictory to this.

5. Limitations of the Study

The present study has few limitations. Firstly, it is a single centre study with a relatively small sample size. Second, our study classified the cases according to activity of UC based on a clinical scoring index- the Truelove and Witts Criteria. This clinical scoring index does not incorporate endoscopic findings to classify cases according to disease activity. Endoscopic findings are the main indicators of mucosal activity. Our study used the Trulove Witts Criteria, as all follow up cases of UC will not be subjected to endoscopy and therefore, there will be a selection bias of enrolling only cases which have undergone endoscopy. To overcome this bias, we utilised the Truelove and Witts Criteria. However, absence of endoscopic correlation is a major drawback.

Third, although CBC is affected by medications such as Azathioprine, Steroids, anti-TNF medications, we did not evaluate the impact of treatment, which could potentially affect the leucocyte count and thus the value of NLR and PLR.

Fourth, our study found a male predominance among cases of UC. However, our study did not investigate the influence of gender on NLR and PLR.

Therefore, to overcome these limitations, multicentric, large cohort, prospective studies are needed and longer follow up periods need to confirm our findings.

6. Conclusion

The present study demonstrates that neutrophil-lymphocyte ratio (NLR) and platelet-lymphocyte ratio (PLR) are simple, inexpensive, and readily available hematological parameters that can be used as useful non-invasive biomarkers for assessing disease activity in patients with ulcerative colitis. Both NLR and PLR were significantly elevated in patients with ulcerative colitis compared to healthy controls, reflecting the underlying inflammatory state.

The study findings suggest that NLR, in particular, showed better overall diagnostic utility compared to PLR, with higher sensitivity and diagnostic accuracy in predicting disease activity. These parameters, being part of routine complete blood count testing, can serve as practical surrogate markers in clinical settings where endoscopic evaluation or fecal biomarkers are not readily available.

Although NLR and PLR cannot replace endoscopy in assessing mucosal healing, they may serve as useful adjunctive tools for disease monitoring and early identification of patients with active or severe disease. Larger multicentric prospective studies with endoscopic correlation are required to validate these findings and establish standardized cut-off values for routine clinical use.

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